**Ward’s Simulated ABO and Rh Blood Typing Lab**

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**Blood Typing Lab**

**Introduction**

How can we tell what blood type a person is? Is there a way to determine whether a sample of blood is RH+ or RH-?

These are the questions we investigated in part one this observational lab. As we are learning about blood types and anti-bodies, a hands-on experiment was the perfect way to exhibit how real blood is classified. We will use “blood” and anti-body serums to learn how to perform a blood typing procedure and indentify the ABO and Rh of our four samples. We will do this by observing the antigen reaction.

In part two, we will take one individual sample of blood to count individual blood cells in the sample. We aim to count the number of blue cells (leukocytes, or white blood cells) and red cells (erythrocytes, or red blood cells).

Through these two processes, we can determine what is necessary in a blood transfusion, and how to classify blood.

**New Vocabulary**

Listed are some key terms that we learned through this lab. In order to understand blood and the process of blood transfusion, it is important to be familiar with these words:

Agglutinogen: an antigen that stimulates the production of an agglutinin  
Agglutinin: an antibody that causes agglutination (clumping of red blood cells)  
Erythrocytes: red blood cells  
Leukocytes: white blood cells

**Materials**

The materials for this lab are as follows:

* 4 blood typing slides (one for each sample person)
* 12 toothpicks (for stirring in order not to mix the blood)
* 1 microscope slide
* 1 cover slip
* 1 compound microscope
* 4 unknown blood samples (Mr. Jones, Mr. Smith, Mr. Green, and Ms. Brown)
* Simulated Anti-A, Anti-B, and Anti-RH serums

**Procedure Part I**

Below, you can see the process of the blood typing experiment:

1. The first step is to set up the lab. To do this, we labeled each blood typing slide with the names of our subjects. (Mr. Jones, Mr. Smith, Mr. Green, and Ms. Brown).
2. We started with Mr. Jones. On slide #1, we placed 3 drops of “blood” onto each of the A, B, and PH wells of the slide.
3. We repeated this step with Mr. Smith in slide #2, Mr. Green in slide # 3, and Ms. Brown in slide #4.
4. Next, we placed 3 drops of anti-A serum in each of the A wells of all four slides. This would show us if the person had type A blood.
5. We repeated this step with the other two wells, adding 3 drops of anti-B serum into the B section of all four slides, and 3 drops of anti-PH into the PH sections.
6. Using the toothpicks, we stirred each well with a clean toothpick for approximately 30 seconds.
7. Finally, we observed each slide to record our results. We were looking for a change from the original – agglutination. We presented this raw data in the charts that follow.

**Procedure Part II**

1. Choose one vial of simulated blood and place one drop on a microscope slide. To properly prepare the microscope slide, place the cover slip on carefully so to avoid air bubbles (which may interfere with the accuracy of results).
2. Place the slide under the microscope and view under low power (10X). Find an area on the slide with a generally even distribution of cells. We used a gridded microscope slide to make it easier to measure the area in which we would count cells.
3. Move the microscope up to high power (40X) and focus the cells again. Then, count the number of red blood cells in the field of view. (The erythrocytes should look similar to red dots.)
4. Next, Count the number of individual simulated white blood cells in the field of view. (The leukocytes should look similar to blue dots).
5. Repeat the previous steps with 2 different fields of views. (Find a different area on the sample to count cells)
6. Using this data, calculate the average number or red and white blood cells (erythrocytes and leukocytes).
7. Finally, multiply the averages by the dilution factor to indentify the number of erythrocytes and leukocytes per cm3. Record the information in a table.

**Raw Data Presentation Part I**

As I experimented on the blood samples, I recorded my raw observations in the charts below. For the first part, we recorded the reaction of the blood to the serum we added – whether or not it was affected by agglutination. By observing the reactions of the blood, we can see which blood type each sample is – we note which anti-body causes a negative reaction.

This chart details the status of the blood when mixed with each different antibody. We also tested the RH factor to see if they were RH positive or negative.

**Part I Table – The ABO and Rh Blood Type of four unknown samples.**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Type | Mr. Jones | Mr. Smith | Mr. Green | Ms. Brown |
| Anti A | Normal – No Agglutination | Agglutination | Agglutination | No reaction |
| Anti B | Color lightened; agglutination | Normal | Cloudy, gel-likes substance – slight agglutination – some reaction | No reaction |
| Anti RH | Normal | Agglutination | Agglutination | No reaction |

As shown, sometimes blood samples react negatively to the serums we added. When a blood sample reacts to an anti serum, we can see what type of blood it is.

**Raw Data Presentation Part II**

In the second part of this lab, we investigated the number of red and white blood cells in a sample of “blood”. As I experimented, my observations and processed data were recorded in the chart below. Our raw data consisted of the data we gathered through the experiment – the number of cells counted in the field of view. Using this data, we generated processed data by calculating the average number of cells counted in the F.O.V (equations listed below) and then using the dilution factor to calculate the number of cells per mm3. The data we gathered from the equations is now processed data. The results of this experiment and the calculations we made show us both the number of cells per mm3 of the sample, and also how the number of erythrocytes and leukocytes compare in number.

**Part II Table - The number of erythrocytes and leukocytes in a blood sample.**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Blood Cell Type** | **Cell Count** | | | **Total # of Cells** | **Avg. # of Cells or Total/3** | **Dilution Factor** | **Total # Blood Cells per mm3** |
| **1** | **2** | **3** |
| **Red** | 104 | 130 | 122 | 356 | 118.67 | 150,000 | 17,800,500 |
| **White** | 6 | 9 | 4 | 19 | 6.33, or 6 1/3 | 5,000 | 31650 |

**Equations used to process data**

Calculating the Avg. # of Red Blood Cells

(Trial 1 + Trial 2 + Trial 3)÷3 = Average # of the three trials  
ex. (104 + 130 + 122)÷3 = 18  
Calculating the Total # of Blood Cells per mm3

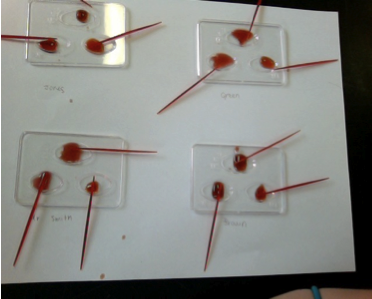
(Avg. # of Red Blood Cells) *×* (Dilution Factor) = Total # of Blood Cells per mm3  
ex. 118.67 *×* 150,000 = 17,800,500

**Visual Evidence from the Experiment**

**Part I**

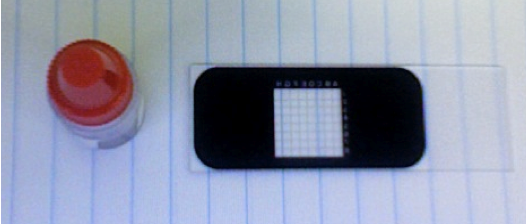


The experiment should be set up in this fashion – each weld should be labeled and contain the blood of an unknown sample.



As shown, once mixed, some blood will show signs of agglutination. By observing these reactions, the blood type of each sample can be identified.

**Part II**



This image shows the process of creating a wet mount. A drop of simulated blood should be added to the slide and a cover slip placed on top. After the slide is prepared, observation can begin.

**Conclusion**

I entered this experiment with a question – what will happen when we add different anti-body serums to our sample blood? Will I be able to determine the blood types of each person? Through careful observations, I came to the following conclusions:

Ms. Brown has the blood type O- and I can see this because, when the serums were added to her blood I saw no reaction at all. This tells me that her blood is universal and is unaffected by any anti-bodies and has no RH factor.

Mr. Green has the blood type of AB+, though I was slightly undecided because of the only slight agglutination from the anti-B serum. I am almost certain, however, that because there was still some noticeable change, his blood resisted the Anti-B.

Mr. Jones has the blood type B- as he had no reaction to the Anti-A or RH serums. This left him as a B-, with agglutination in the Anti-B sample.

Mr. Smith has the blood type A+ as he shows agglutination with the Anti-A and Anti-Rh serum.

This lab clearly demonstrated the process and evaluation of blood typing.

In part two of this lab, the evidence shows that there are far more erythrocytes than leukocytes. Through our observation, the data showed that, on average, there was a higher number of red blood cells and, in most cases, a relatively low number of white blood cells. This gives an insight about the structure of blood and the function of each type of cell - in normal blood, it is necessary to have a higher number of red blood cells. Also, using different equations, we formulated our raw data and numbers into a conclusion – the amount of each type of cell per mm3. The number of red blood cells is about 500x the number of white blood cells.

**Graph to Demonstrate Data**

The following graph visually represents the data we collected and is evidence for my conclusion. The number of white blood cells is less than 1% of the total while the red blood cells make up 99%. This shows the vast difference in amount between the erythrocytes and leukocytes.

As shown, the percentage of white blood cells is tremendously lower than that of the red blood cells.

**Evaluation**

Looking back on the experiment, I found many strengths and weaknesses in the procedure. In terms of strengths, time was managed well and the calculations were performed accurately. Also, I followed the instructions carefully, made precise observations, and understood how to draw conclusions from my results. A weakness was the difficulty and possible inaccuracy of the cell counting. The sheer number of cells and the fact that many overlapped made it difficult to get the exact number. However, the fact that we did 3 trials and calculated the averages allowed for a more accurate conclusion and smaller margin for error.

An improvement to the experiment would be to limit the number of air bubbles in the wet mount as these often interfered with my ability to count the cells accurately. Also, I had difficulty seeing the difference between the red dots and blue dots as I could only barely see them in the high power. Under the maximum power, it was easy to distinguish the different colors, but hard to track the field of view. For this reason, I decided to stay in the 40X power (which may have caused a slight error or discrepancy in my results). Regardless, the overall experiment allowed me to really understand this process and understand key procedures in experimenting. I also came to accurate conclusions through my data and learned how to interpret my results.